



USSN: 09/551,977  
Dkt. No.: PP001593.0004  
2300-1593

AF 3m

**PATENT**

**CERTIFICATE OF MAILING PURSUANT TO 37 CFR § 1.8**

I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to: Mail Stop Appeal Brief, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450 on **May 24, 2006**.

5/24/06

Date

Michelle Hobson

Signature

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

In Re Application of:

POLO et al.

Serial No.: 09/551,977

Filing Date: April 14, 2000

Title: COMPOSITIONS AND METHODS FOR  
GENERATING AN IMMUNE RESPONSE  
UTILIZING ALPHAVIRUS-BASED  
VECTOR SYSTEMS

Examiner: B. Li

Group Art Unit: 1648

Confirmation No.: 2230

Customer No.:

**TRANSMITTAL LETTER**

Mail Stop Appeal Brief  
Commissioner for Patents  
P.O. Box 1450  
Alexandria, VA 22313

Sir:

Transmitted herewith for filing, please find the following documents:

- Reply Brief (14 pgs) with attached Claims Appendix (2 pgs) and Evidence Appendix (5 pages)
- Return receipt postcard

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The fee is calculated as follows:

	NO. OF CLAIMS	CLAIMS PREVIOUSLY PAID FOR	EXTRA CLAIMS	RATE	FEE
Total Claims	6	- 37	0	x \$50.00	\$0
Independent Claims	1	- 9	0	x \$200.00	\$0
Multiple dependent claims not previously presented, add \$360.00					\$0
Total Amendment Fee					\$0
Petition for Extension of Time Fee					\$0
Small Entity Reduction (if applicable)					\$0
<b>TOTAL FEE DUE</b>					<b>\$0</b>

The Commissioner is hereby authorized to charge any appropriate fees under 37 C.F.R. §§1.16, 1.17, and 1.21 that may be required by this paper, and to credit any overpayment, to Deposit Account No. 18-1648.

Respectfully submitted,

Date: May 24, 2006

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**REPLY BRIEF**

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**REPLY BRIEF**

Mail Stop Appeal Brief  
Commissioner for Patents  
Alexandria, VA 22313

Sir:

Pursuant to Section 41.37(c) (69 Fed. Reg. 49962, Aug 2004), Applicants submit the following Reply Brief in Response to the second Examiner's Answer mailed on March 24, 2006. This Reply Brief is submitted within two months of the date of mailing of the second Examiner's Answer, namely by May 24, 2006. Appellants respectfully request that the decision of the Examiner be reversed.

### **STATUS OF THE CLAIMS**

Claims 17 and 19-23 are currently pending in the above-referenced case (hereinafter "the application"). Claim 20 is allowable and claims 17, 19 and 21-23 remain rejected under 35 U.S.C. § 112, first paragraph (written description).

### **GROUNDS OF REJECTION**

1. Claims 17, 19 and 21-23 stand rejected under 35 U.S.C. § 112, 1<sup>st</sup> paragraph as not being adequately described by the specification as filed.

### **ARGUMENTS**

#### **1. The Specification Describes the *Claimed* Subject Matter**

Appellants incorporate herein all the arguments set forth in their Appeal Brief and first Reply Brief, filed December 5, 2005. Rather than reiterate each argument herein, Appellants address various assertions set forth in the second Examiner's Answer, in which it was again maintained that the specification as filed does not adequately describe claims 17, 19 and 21-23. Appellants address the points raised in the second Examiner's Answer in turn.

##### **(a) The Examiner Has Misconstrued the *Claimed* Subject Matter**

Appellants reiterate that the Examiner's rejection is based on an improper construction of the claimed subject matter. Throughout prosecution the Examiner has insisted that, in order to satisfy the written description requirement, the as-filed specification must set forth both the particular amino acid residues in positions 158-162 of any given alphavirus's E2 protein and possible mutations at this positions.<sup>1</sup>

However, Applicants are not claiming recombinant alphavirus particles in which residues 158-162 of the E2 are mutated, regardless of the alphavirus from which the E2 protein is

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<sup>1</sup> See, e.g., second Examiner's Answer, pages 6-7

obtained. Rather, the claims are drawn to a recombinant alphavirus particle having “a mutation in the E2 glycoprotein is in the region corresponding to amino acids 158 - 162, numbered relative to wild-type SIN E2 glycoprotein” and which recombinant particle infects human dendritic cells. In other words, contrary to the Examiner’s assertions, the mutations(s) in E2 required by claim language is(are) not at whichever residues are at positions 158-162 of that E2 protein but, instead, are determined relative to a reference sequence, namely Sindbis E2.

In addition, Appellants reiterate that the claims also contain a functional limitation that the Examiner has repeatedly failed to take into consideration. Not only are, at most, 5 amino acids of an alphavirus E2 protein altered, but, in addition, that altered E2 protein, when incorporated into a recombinant alphavirus particle, must result in a particle that is capable of infecting human dendritic cells. This functional limitation is clearly described in the as-filed specification<sup>2</sup> and, moreover, further limits the genus of the claimed molecules in a way that the Examiner has failed to address.

When the claims are properly construed, *i.e.*, so that the position of the mutations in E2 are determined *relative to* Sindbis E2 and functional limitations considered, it is clear that the length of a particular alphavirus E2 protein, the particular residues at positions 158-162 of any given alphavirus E2 protein<sup>3</sup> and even the particular residues *corresponding to* residues 158-162 of wild type SIN E2 need not be set forth in the as-filed specification in order to satisfy the written description. The relevant inquiry is whether the specification and/or art describes how to determine which residues of **any** alphavirus E2 protein correspond to amino acids 158-162 of E2 of Sindbis so that one or more of these residues can be altered and a recombinant alphavirus particle including the mutated E2 protein can be tested for human dendritic cell infectivity.

The evidence of record clearly establishes that aligning E2 proteins and testing recombinant alphavirus particles for dendritic cell infectivity were well known in the art and,

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<sup>2</sup> See, e.g., page 19, line 18 to page 20, line 5

<sup>3</sup> See, attachments of GenBank Accession Nos and page 6 of second Examiner’s Answer setting forth amino acid residues at positions 158-162 of various E2 alphavirus proteins

accordingly, need not be described in the specification.<sup>4</sup> Furthermore, although not required, the relevant claimed features are also described in the as-filed specification.<sup>5</sup>

Still further evidence of the ample description regarding determining the 5 amino acid region of an alphavirus E2 protein that contains the mutation(s) is submitted herewith. In particular, attached hereto is an alignment of various alphavirus E2 proteins. It is readily apparent that this (and other) alignments clearly show that, following the clear description in the specification and known methods, the skilled artisan would have known that Appellants were in possession of any recombinant alphavirus particle having a mutation corresponding to positions 158-162 (LKETS) of a Sindbis E2 protein.<sup>6</sup> Because the targeted residues are identified in relation to a particular sequence (SIN E2) and because determining which residues correspond to the reference sequences was described and well known, satisfaction of the written description requirement cannot require that the as-filed specification set forth particular residues at these positions in any given alphavirus.

In sum, the region corresponding to 158-162 of SIN E2 can be readily determined for **any** alphavirus, regardless of the length or particular sequence of the E2 protein.<sup>7</sup> Similarly, the specification clearly describes how to assay a recombinant alphavirus particle containing the recited mutation(s) for human dendritic cell infectivity. Accordingly, satisfaction of the written description requirement has been established by showing that Appellants were in possession of **any** recombinant alphavirus particle having one or more mutations in specified 5-amino acid region and which particle infects human dendritic cells.

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<sup>4</sup> See, e.g., *Capon v. Eshhar* 76 USPQ2d 1078 (CA FC 2005)

<sup>5</sup> See,, e.g., page 5, lines 4-6 (describing mutation in E2 corresponding to residues 158-162 of Sindbis); page 5, lines 7-8 (describing deletions or insertions in this identified region); page 5, lines 9-10 (describing various alphavirus types)

<sup>6</sup> Evidence Appendix A, aligning E2 proteins from various alphaviruses (see, also, page 1 of the specification)

<sup>7</sup> Appellants also traverse the Examiner's assertions on page 7 of the second Examiner's Answer that 41 alphavirus species constitute an "innumerable" number of species that render the claims very broad. Aligning any of these 41 alphavirus species to Sindbis is more than adequately described

**(b) A Core Region Common to All Alphaviruses is Described in the As-Filed Specification**

Appellants also traverse the Examiner's repeated assertion that the specification does not adequately describe "which position corresponds to amino acids 158-162 of Sindbis virus E2 that is important for the amino acid substitutive mutation."<sup>8</sup>

As a threshold matter, Appellants again note that, by definition, a mutation in a virus's envelope glycoprotein that alters cell tropism does so by changing the way the envelope glycoprotein interacts with the viral receptor of a cell. This is because only short stretches of amino acid residues (*e.g.*, loops) exposed on the surface of the envelope glycoprotein make contact with the viral receptor on the cell. It was known at the time of filing that mutations within these surface-exposed regions can give rise to the altered receptor binding and, accordingly, altered cell tropism. In the pending case, for the reasons of record and reiterated herein, the specification clearly describes the importance of the 158-162 region of Sindbis virus E2 for conferring human dendritic cell tropism and, as such, mutations at any of the positions corresponding to positions 158-162 of Sindbis virus E2 are supported by the description.

The evidence of record (including Evidence Appendix attached hereto) clearly establishes that there is ample description of the claimed alphavirus particles, including identification of a 5 amino acid region which is the subject of mutation. This 5 amino acid region that includes the mutation(s) is a small fraction of any alphavirus E2 protein. As acknowledged by the Examiner, alphavirus E2 proteins are at least 420 amino acid residues in length.<sup>9</sup> Thus, at most, the claimed particles contain mutations in approximately 1% of an E2 protein.

In any event, the evidence demonstrates that all alphaviruses contain an E2 protein and that the length of the particular E2 protein is irrelevant because it is both described in the specification and well known how to determine which residues correspond to the specified 158-

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<sup>8</sup> Second Examiner's Answer, page 9

<sup>9</sup> Second Examiner's Answer, page 6

162 residues of SIN E2. Thus, contrary to the Examiner's assertion, a defined and small core region is clearly described in the as-filed specification as the region subject to mutation.

**(c) A Single Species Can Adequately Describe a Genus**

As previously, the second Examiner's Answer also asserts that "the evidence indicates that an ordinary artisan could not predict the operability in the invention of any species other than that disclosed."<sup>10</sup> In addition, although the Examiner admitted that the specification describes how to test "the mutated alphaviruses for human dendritic cell tropism,"<sup>11</sup> it was alleged that these teachings are not relevant to determining adequacy of written description.

Contrary to the Examiner's assertions, it is well settled that disclosure of a single species in combination with disclosure of an assay for testing functional limitations can provide an adequate description for a broad genus.<sup>12</sup> In particular, the PTO Guidelines<sup>13</sup>, favorably commented on by the Federal Circuit in *Enzo Biochem Inc. v. Gen-Probe Inc.*, 63 USPQ2d 1609, 1617 (Fed. Cir. 2003), include various Examples that establish that claims to a genus of molecules is properly described if (1) the claimed molecule is novel, (2) unobvious, (3) a specific activity is recited; and (4) assays for identifying all variants having the claimed activity are provided:

The single species disclosed is representative of the genus because all members have at least 95% structural identity with the reference compound and because of the presence of an assay which applicant provided for identifying all of the at least 95% identical variants of SEQ ID NO:3 which are capable of the specified catalytic activity. One of skill in the art would conclude that applicant was in

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<sup>10</sup> Second Examiner's Answer, page 11, citing *Enzo Biochem*

<sup>11</sup> Second Examiner's Answer, page 11

<sup>12</sup> See, Appellants Appeal Brief and first Reply Brief

<sup>13</sup> See, e.g., Examples 9 and 14 of PTO Guidelines, reproduced in part in Appellants' Appeal Brief

possession of the necessary common attributes possessed by the members of the genus.<sup>14</sup>

Therefore, in the case on appeal, the Examiner errs in asserting that Appellants have only described a recombinant alphavirus particle containing the particular exemplified E2 mutant. In addition, the Examiner has also errs in asserting that the assays for testing human dendritic cell tropism disclosed in as-filed specification assays are not relevant to the written description inquiry.

The test for determining satisfaction of the requirement of Section 112, 1<sup>st</sup> paragraph is not what molecules are actually described (or reduced to practice) in the as-filed specification, but, rather, what the disclosure as a whole and available knowledge to determine whether the specification as-filed evinces possession of the claimed subject matter to the skilled artisan. In light of the disclosure of a representative species having the claimed structural (mutation in one of 5 amino acid core region) and functional (human dendritic cell tropism) features, along with an assay for determining all other variants of the genus, the skilled artisan would have no doubts that Appellants were in possession of the claimed alphavirus particles at the time of filing.

Therefore, for the reasons of record and those set forth herein, the as-filed specification more than satisfies the written description requirement of 35 U.S.C. § 112, 1<sup>st</sup> paragraph.

**(d) The Cases Cited in the Examiner’s Answer Support a Determination that the Written Description Requirement Has Been Satisfied**

With regard to the various cases cited in the Examiner’s Answer,<sup>15</sup> Appellants again note that the holding shared by all written description cases is that the written description inquiry is fact-dependent. Thus, satisfaction of the written description requirement will depend on what is claimed. Claims drawn to molecules that having particular functions (*e.g.*, enzymes in Example 14 above, infection of human dendritic cells as in the case on appeal) may require that the specification describe a core region associated with the claimed function. As established in

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<sup>14</sup> See, Example 14 of PTO Guidelines entitled “Product-by-Function”

<sup>15</sup> Second Examiner’s Answer, pages 10-11

Example 14 of the PTO Guidelines, this does not mean that every possible amino acid substitution in the identified “core” region needs to described or tested. Instead, the description of a single species coupled with the description of an assay for determining the other methods of genus is more than sufficient to satisfy the written description requirement. Based on the evidence of record, Appellants have more than amply satisfied this requirement.

Indeed, Appellants reiterate that the specification at issue in every case cited by the Examiner did not contain a literal description of the claimed subject matter or of a core region. In contrast, Appellants’ specification contains express support for the claimed invention and, accordingly, possession of the invention at the time the application was filed has been established.

The cases cited by the Examiner also affirm the well-established rule that an applicant need not describe that which is not new. As set forth in *Capon v. Eshhar* 76 USPQ2d 1078 (Fed. Cir. 2005), the Federal Circuit rejected the notion that the specification must describe information (e.g., sequence data) that is either known or can readily be determined based on scientific facts (*Capon* at page 1085, emphasis added):

It is not necessary that every permutation within a generally operable invention be effective in order for an inventor to obtain a generic claim, provided that the effect is sufficiently demonstrated to characterize a generic invention. See *In re Angstadt*, 537 F.2d 498, 504 [190 USPQ 214] (CCPA 1976) (“The examples, both operative and inoperative, are the best guidance this art permits, as far as we can conclude from the record”). While the Board is correct that a generic invention requires adequate support, the sufficiency of the support must be determined in the particular case. ...

As in *Capon*, the Examiner’s assertion in the instant case that Appellants are required to disclose particular mutations in each and every alphavirus species described in the specification, when the E2 sequences of these (and other alphaviruses) were well known and could be readily aligned with the prototype SIN to determine residues corresponding to 158-162, is inappropriate.

Appellants have already identified the structure-function correlation by pointing to a region of 5 amino acids that is mutated and by reciting that the mutated molecule must have DC infectivity.

Moreover, the Examiner admits that it was well known that (1) all alphaviruses contain E2; (2) alphavirus E2 proteins were well characterized; (3) aligning amino acid sequences was routine (and literally described by the specification as filed with respect to alphaviruses, for example on page 37); and (4) assaying for the claimed function limitation was well known (and literally described by the specification as filed). Combined with the express disclosure of the 5 particular amino acids subject to mutation, the skilled artisan would have recognized that Appellants were in possession of the claimed genus of recombinant alphavirus particles at the time the present application was filed.

**(e) Dr. Polo's Declaration Establishes the Adequacy of Written Description for the Claimed Molecules**

In the Examiner's Answer, it was asserted that Dr. Polo's Declaration was considered but was not persuasive to overcome of the "enablement" rejection.<sup>16</sup> Inasmuch as this Declaration was already found by the Examiner to overcome the previous enablement rejection, Appellants assume the Examiner was referring to the outstanding written description rejection.

In any event, Appellants reiterate that Dr. Polo's Declaration directly addresses written description issues, for example in paragraphs 7, 8 and 11, emphasis added:

7. When the specification was filed, it clearly taught a typical scientist how to make and use recombinant alphavirus particles from a variety of alphavirus species, where the particles are capable of infecting human dendritic cells and contain an amino acid mutation at positions 158-162 (based on SIN numbering) of E2 (relative to the wild-type alphavirus source). **Thus, I believe that a typical scientist would have understood the specification clearly described all of the various aspects of the claims and enabled a typical scientist to make and use the invention as set forth in the pending claims.** I base this belief on the facts set forth below.

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<sup>16</sup> Second Examiner's Answer, page 12

8. ... In view of the teachings of the specification, it would have been routine for the skilled artisan to align and compare nucleotide and amino acid sequences from various alphaviruses and determine which amino acid sequences in any alphavirus corresponded to positions 158-162 of a SIN E2 protein. (See, e.g., page 37 of the specification, **describing** alignment of SIN strains). Also, in view of the **disclosure**, a person of skill in the art would surmise that mutants in this region would be much more likely to exhibit DC-tropism. Accordingly, it is my opinion that using the teachings of the specification and state of the art, it would require only routine experimentation for a typical scientist to obtain suitable amino acid sequences from any alphavirus (for example by comparison with sequences disclosed in the specification) and use these alphavirus sequences as a starting point for making the claimed particles.

11. Fourth, it would have been clear to a typical scientist how to test for the ability of a mutant alphavirus particle falling within the scope of the claims to infect human dendritic cells. (See, e.g., page 42 of the specification). Methods of culturing human dendritic cells were **known and described** in the specification as filed. (See, e.g., Example 1). Moreover, methods of testing the ability of alphavirus particle to infect these cells **are described** in detail in the specification and include, but are not limited to, testing FACS analysis, titer analysis, use of reporter molecules, and the like. (See, e.g., page 40; page 42-43). Thus, it is my opinion that a typical scientist could have readily tested any recombinant alphavirus particle containing the claimed mutation, following the teachings of the specification.

Disclosure of a single representative species along with disclosure of assays for determining other representative species evinces possession of a genus. Therefore, Appellants again submit that Dr. Polo's Declaration, which establishes that Appellants describe a representative SIN species as well as assays for determining human DC-infectivity, directly addresses written description and establishes that Appellants were in possession of the claimed particles at the time of filing.

In summary, for the reasons of record as set forth herein, in the Appeal Brief, and in the first Reply Brief, the written description rejection cannot stand. As would be recognized by the skilled artisan, the originally filed disclosure describes all features of the claimed recombinant alphavirus particles which infect human dendritic cells. Thus, the specification sufficiently describes the claimed invention and Appellants have satisfied the written description requirement.

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**CONCLUSION**

For the reasons stated above, Appellants respectfully submit that the pending claims are described by the specification as filed. Accordingly, Appellants request that the rejection of the claims on appeal be reversed, and that the application be remanded to the Examiner so that the appealed claims can proceed to allowance.

Respectfully submitted,

Date: May 24, 2006

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## CLAIMS APPENDIX



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## **CLAIMS INVOLVED IN THE APPEAL**

1 to 16. (canceled).

17. (previously presented): A recombinant alphavirus particle comprising an alphavirus replicon comprising a heterologous sequence; and an amino acid mutation in its E2 glycoprotein, wherein the mutation in the E2 glycoprotein is in the region corresponding to amino acids 158 - 162, numbered relative to wild-type SIN E2 glycoprotein, and further wherein said particle is capable of infecting human dendritic cells, with the proviso that said recombinant alphavirus particle is not derived from ATCC # VR-2526.

18. (canceled).

19. (previously presented): The recombinant alphavirus particle of claim 17 wherein said alphavirus is a Sindbis virus.

20. (original): The recombinant alphavirus particle according to claim 19 wherein said alphavirus has an amino acid substitution at E2 residue 160, as compared to wild-type Sindbis virus.

21. (previously presented): The recombinant alphavirus particle according to claim 17 wherein said alphavirus is Semliki Forest virus.

22. (previously presented): The recombinant alphavirus particle according to claim 17 wherein said alphavirus is Ross River virus.

23. (previously presented): The recombinant alphavirus particle according to claim 17 wherein said alphavirus is Venezuelan equine encephalitis virus.

24 to 37. (canceled).

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## EVIDENCE APPENDIX

The Evidence Appendix consists of one (1) document showing an alignment of E2 proteins from various alphavirus (Sindbis, Western encephalitis, Eastern equine encephalitis, Semliki forest virus, Venezuelan equine encephalitis, Barmah, Middleburg and Ndumu virus). This Appendix is submitted for the first time herewith in response to the GenBank sequences attached to the Examiner's Answer that had not been previously cited by the Examiner.

Thursday, March 16, 2006 4:12 PM

	S	V	T	E	H	F	T	A	Y	K	L	R	P	Y	I	A	Y	C	P	D	C	G	H	G	E	S	C	H	S	Majority	
1	S	V	T	G	D	F	T	-	-	L	T	S	P	Y	L	G	T	C	P	Y	C	H	H	T	E	P	C	F	S	wt sine2	
1	S	I	T	D	D	F	T	-	-	L	T	S	P	Y	L	G	F	C	P	Y	C	R	H	S	T	P	C	F	S	Western equine	
1	D	L	D	T	H	F	T	Q	Y	K	L	A	R	P	Y	I	A	D	C	P	N	C	G	H	S	-	R	C	D	S	Eastern equine
1	S	V	S	Q	H	F	N	V	Y	K	A	T	R	P	Y	I	A	Y	C	A	D	C	G	A	G	H	S	C	H	S	Semliki E2
1	S	T	E	E	L	F	K	E	Y	K	L	T	R	P	Y	M	A	R	C	I	R	C	A	V	G	-	S	C	H	S	venezuelan
1	S	V	A	-	H	F	E	A	Y	K	A	T	R	P	Y	I	G	W	C	A	D	C	G	L	A	G	S	C	P	S	Barmah
1	G	L	T	E	D	Y	K	A	Y	K	L	T	K	P	Y	I	A	Y	C	S	D	C	G	N	G	Q	F	C	Y	S	middleburg
1	A	-	A	Q	H	F	N	E	Y	K	V	T	Q	P	Y	I	A	Y	C	P	D	C	G	N	G	E	S	C	Q	S	ndumu
	P	I	A	I	E	A	V	R	S	E	A	D	D	G	V	L	K	I	Q	T	S	A	Q	I	G	L	D	K	S	G	Majority
28	P	I	K	I	E	Q	V	W	D	E	P	D	D	T	T	I	R	I	Q	T	S	A	Q	F	G	Y	D	Q	S	G	wt sine2
28	P	I	K	I	E	N	V	W	D	E	S	D	D	G	S	I	R	I	Q	V	S	A	Q	F	G	Y	N	Q	A	G	Western equine
30	P	I	A	I	E	E	V	R	G	D	A	H	A	G	V	I	R	I	Q	T	S	A	M	F	G	L	K	T	D	G	Eastern equine
31	P	V	A	I	E	A	V	R	S	E	A	T	D	G	M	L	K	I	Q	F	S	A	Q	I	G	I	D	K	S	D	Semliki E2
30	P	I	A	I	E	A	V	K	S	D	G	H	D	G	Y	V	R	L	Q	T	S	S	Q	Y	G	L	D	S	S	G	venezuelan
30	P	V	S	I	E	H	V	W	S	D	A	D	D	G	V	L	K	I	Q	V	S	M	Q	I	G	I	A	K	S	N	Barmah
31	P	I	A	I	E	R	V	R	A	E	A	S	D	G	M	L	K	I	Q	I	S	A	Q	I	G	L	Q	V	D	G	middleburg
30	P	V	A	I	E	R	I	R	N	E	A	T	D	G	T	L	K	V	Q	F	A	L	Q	V	G	I	K	K	D	H	ndumu
	A	T	D	V	T	K	I	R	Y	M	S	-	-	G	H	T	V	K	E	A	X	R	S	S	L	S	V	S	T	Majority	
58	A	T	S	V	N	K	Y	R	Y	M	S	F	D	Q	D	H	T	V	K	E	G	Q	M	D	D	I	K	I	S	T	wt sine2
58	T	A	D	V	T	K	F	R	Y	M	S	F	D	H	D	H	D	I	K	E	D	S	M	E	K	I	A	I	S	Western equine	
60	-	V	D	L	-	-	A	Y	M	S	F	M	N	G	K	T	Q	K	S	I	K	I	D	N	L	H	V	R	T	Eastern equine	
61	N	H	D	Y	T	K	I	R	Y	A	D	-	-	G	H	A	I	E	N	A	V	R	S	S	L	K	V	A	T	Semliki E2	
60	N	L	K	G	-	-	R	T	M	R	Y	D	M	H	G	T	I	E	E	I	P	L	H	Q	V	S	L	H	T	venezuelan	
60	T	I	N	H	A	K	I	R	Y	M	G	-	-	A	N	G	V	Q	E	A	R	S	T	L	S	V	S	T	Barmah		
61	A	H	S	W	T	K	I	R	Y	M	K	-	-	G	H	D	V	E	D	T	D	R	N	S	L	E	V	F	T	middleburg	
60	S	T	A	P	T	Y	L	R	Y	M	E	-	-	G	H	E	V	K	D	A	L	-	N	N	L	R	V	Y	T	ndumu	
	S	G	P	C	R	V	L	G	T	M	G	H	F	I	L	A	R	C	P	P	G	D	S	V	T	V	S	F	V	D	Majority
100	S	G	P	C	R	R	L	G	H	K	G	Y	F	L	L	A	K	C	P	P	G	D	S	V	T	V	S	I	V	S	wt sine2
88	S	G	P	C	R	R	L	G	H	K	G	Y	F	L	L	A	Q	C	P	P	G	D	S	V	T	V	S	I	T	S	Western equine
86	S	A	P	C	S	L	V	S	H	H	G	Y	Y	I	L	A	Q	C	P	P	G	D	T	V	T	V	G	F	H	D	Eastern equine
88	S	G	D	C	F	V	H	G	T	M	G	H	F	I	L	A	R	C	P	P	G	E	F	L	Q	V	S	I	Q	D	Semliki E2
87	S	R	P	C	H	I	V	D	G	H	G	Y	F	L	L	A	R	C	P	A	G	D	S	I	T	M	E	F	K	K	venezuelan
87	T	A	P	C	D	I	L	A	T	M	G	H	F	I	L	A	R	C	R	P	G	S	Q	V	E	V	S	L	S	T	Barmah
88	T	G	E	C	T	V	H	G	T	M	G	H	F	I	V	A	T	C	P	E	G	D	S	L	T	V	A	F	V	D	middleburg
86	S	A	Q	C	E	V	T	A	T	M	G	H	F	A	L	A	R	C	P	P	G	D	S	L	S	V	T	F	M	D	ndumu

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		Majority																	
		130						140						150					
118	S S S - T T S C T L A R K I K P K F V G R E R Y D L P P V Y	wt sine2																	
118	G A S - E N S C T V E K K I R R K F V G R E E Y L F P P V H	Western equine																	
116	G P N - R H T C T V A H K V E F R P V G R E K Y R H P P E H	Eastern equine																	
118	T R N A V R A C R I Q Y H H D P Q P V G R E K F T I R P H Y	Semliki E2																	
117	G S V - T H S C S V P Y E V K F N P V G R E L Y T H P P E H	venezuelan																	
117	D P K L - - L C R T P F S H K P R F I G N E K S P A P T G H	Barmah																	
118	K H K V R H A C R I A Y K H R V P V L G R E H F T V R P H H	middleburg																	
116	A S K K N R M C R V P F V H K L P F L G R E K N S A R R Y H	ndumu																	
		Majority																	
		160						170						180					
147	G K N I P C R M Y D R L K E T S A G Y I T M H R P G P H A Y	wt sine2																	
147	G K L V K C H V Y D H L K E T S A G Y I T M H R P G P H A Y	Western equine																	
145	G V E L P C N R Y T H K R A D Q G H Y V E M H Q P G L V A D	Eastern equine																	
148	G K E I P C T T Y Q Q T T A E T V E E I D M H M P P D T P D	Semliki E2																	
146	G A E Q A C Q V Y A H D A Q N R G A Y V E M H L P G S E V D	venezuelan																	
145	K T R I P C K T Y S H Q T D L T R E E I T M H V P P D V P I	Barmah																	
148	G V E L P C T T Y A M R T S V T T E E I E M H V A H D V P D	middleburg																	
146	G K D V T C T T Y A R R T D V T D E N L E M H V P P H I P D	ndumu																	
		Majority																	
		190						200						210					
177	T S Y L E E A S G K I Y A K P P S G K N I T Y E C K C G D Y	wt sine2																	
177	K S Y L E E A S G E V Y I K P P S G K N V T Y E C K C G D Y	Western equine																	
175	H S L L S I H S A K V K I T V P S G A Q V K Y Y C K C P D V	Eastern equine																	
178	R T L L S Q Q S G N V K I T V - G G K K V K Y N C T C - G T	Semliki E2																	
176	S S L I S L S G S S V T V T P P V G T S A L V K C K C G G T	venezuelan																	
175	Q G L V S N T G K S Y S L D - P K T K T I K Y K C T C G E T	Barmah																	
178	N T F L S K T G N K V K I T P - K G K S I R Y N C T C - G S	middleburg																	
176	A T L V K V Q L N N V T I T P P S G A T V R Y N C S C K D G	ndumu																	
		Majority																	
		220						230						240					
207	K T G T V K T R - T E I T G C T A I K Q C V A Y K S D Q T K	wt sine2																	
207	S T G I V S T R - T K M N G C T K A K Q C I A Y K S D Q T K	Western equine																	
205	R E G T T S S D Y T - T T C T D V K Q C R A Y L I D N K K	Eastern equine																	
206	G N V G T T N S D M T I N T C L I E - Q C H V S V T D H K K	Semliki E2																	
206	K I S E T I N K A K Q F S Q C T K K E Q C R A Y R L Q N D K	venezuelan																	
204	V K E G T A T N K I T L F N C D T A P K C I T Y A V D N T V	Barmah																	
206	K E S G V T K Q D K E F D N C E V S - Q C H T M V T A H D K	middleburg																	
206	N R Q S E T S R E V V L S G C A E A - K C H A A V V D G K V	ndumu																	

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	W	V	F	N	S	P	D	V	P	R	A	E	G	T	A	V	K	G	K	L	H	V	P	F	P	L	T	N	S	T	Majority
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250

260

270

236	W	V	F	N	S	P	D	L	I	R	H	A	D	H	A	A	Q	G	K	L	H	L	P	F	R	L	V	P	S	S	wt sine2
236	W	V	F	N	S	P	D	L	I	R	H	T	D	H	S	V	Q	G	K	L	H	I	P	F	R	L	T	P	T	V	Western equine
233	W	V	Y	N	S	G	R	L	P	R	G	E	G	D	T	F	K	G	K	L	H	V	P	F	V	P	V	A	K	Eastern equine	
235	W	Q	F	N	S	P	F	V	P	R	A	D	E	P	A	R	K	G	K	V	H	I	P	F	P	L	D	N	I	T	Semliki E2
236	W	V	Y	N	S	D	K	L	P	K	A	A	G	A	T	L	K	G	K	L	H	V	P	F	L	L	A	D	G	K	venezuelan
234	W	Q	Y	N	S	Q	Y	V	P	R	S	E	V	T	E	V	K	G	K	I	H	V	P	F	P	L	T	D	S	T	Barmah
235	W	Q	F	N	S	P	Y	V	P	R	A	G	-	S	G	K	K	G	K	I	H	V	P	F	P	L	S	N	S	T	middleburg
235	W	Q	Y	S	S	R	F	V	P	R	H	E	K	T	E	L	K	G	K	V	H	V	P	F	P	L	T	N	T	T	ndumu

	C	R	V	P	L	A	P	E	P	T	V	T	H	G	F	R	G	V	T	L	H	L	H	P	D	H	P	T	L	L	Majority
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280

290

300

266	C	K	V	P	V	A	H	A	P	S	V	V	H	G	F	K	H	I	S	L	Q	L	D	T	D	H	L	T	L	L	wt sine2
266	C	P	V	P	L	A	H	T	P	T	V	T	K	W	F	K	G	I	T	L	H	L	T	A	M	R	P	T	L	L	Western equine
263	C	I	A	T	L	A	P	E	P	L	V	E	H	K	H	R	T	L	I	L	H	L	Y	P	D	H	P	T	L	L	Eastern equine
265	C	R	V	P	M	A	R	E	P	T	V	I	H	G	K	R	E	V	T	L	H	L	H	P	D	H	P	T	L	F	Semliki E2
266	C	T	V	P	L	A	P	E	P	M	I	T	F	G	F	R	S	V	S	L	K	L	H	P	K	N	P	T	Y	L	venezuelan
264	C	A	V	S	V	A	P	E	P	Q	V	T	Y	R	L	G	E	V	E	F	H	F	H	P	M	Y	P	T	L	F	Barmah
264	C	R	V	P	L	A	P	L	P	N	T	I	P	A	K	N	G	I	T	L	Q	L	H	P	V	A	P	T	L	L	middleburg
265	C	R	V	S	V	A	T	K	P	I	E	K	H	A	R	G	L	L	T	L	V	L	H	P	E	Y	P	T	L	L	ndumu

	T	T	R	S	L	G	A	D	P	E	P	T	X	E	W	I	S	E	P	T	E	R	N	F	P	V	G	E	E	G	Majority
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310

320

330

296	T	T	R	R	L	G	A	N	P	E	P	T	S	E	W	I	I	G	K	T	V	R	N	F	S	V	G	R	D	G	wt sine2
296	T	T	R	K	L	G	L	R	A	D	A	T	A	E	W	I	T	G	S	T	S	R	N	F	S	V	G	R	E	G	Western equine
293	T	T	R	S	L	G	S	D	A	N	P	T	R	Q	W	I	E	R	P	T	T	V	N	F	T	V	T	G	E	G	Eastern equine
295	S	Y	R	T	L	G	E	D	P	Q	Y	H	E	E	W	V	T	A	A	V	E	R	T	I	P	V	P	V	D	G	Semliki E2
296	T	T	R	Q	L	A	D	E	P	H	Y	T	H	E	L	I	S	E	P	A	V	R	N	F	T	V	T	E	K	G	venezuelan
294	S	I	R	S	L	G	K	D	P	S	H	S	Q	E	W	I	D	T	P	M	S	K	T	I	Q	V	G	A	E	G	Barmah
294	T	Y	R	T	L	G	E	K	P	E	H	H	T	E	W	I	S	E	S	C	E	R	T	L	P	V	P	E	E	G	middleburg
295	S	Y	R	Y	L	G	A	E	P	R	P	V	N	R	W	I	S	E	P	T	E	V	Q	I	P	V	S	K	E	G	ndumu

	L	E	Y	V	W	G	N	H	D	P	V	R	V	W	A	Q	E	S	A	P	G	S	P	H	G	W	P	H	E	I	Majority
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340

350

360

326	L	E	Y	T	W	G	N	H	D	P	V	R	V	Y	A	Q	E	S	A	P	G	D	P	H	G	W	P	H	E	I	wt sine2
326	L	E	Y	V	W	G	N	H	E	P	V	R	V	W	A	Q	E	S	A	P	G	D	P	H	G	W	P	H	E	I	Western equine
323	L	E	Y	T	W	G	N	H	P	P	K	R	V	W	A	Q	E	S	G	E	G	N	P	H	G	W	P	H	E	V	Eastern equine
325	M	E	Y	H	W	G	N	N	D	P	V	R	L	W	S	Q	L	T	T	E	G	K	P	H	G	W	P	H	Q	I	Semliki E2
326	W	E	F	V	W	G	N	H	P	P	K	R	F	W	A																

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I S H Y Y H R Y P A T T V L V L V G A A A A I L V S L A A S Majority

370

380

390

356	I Q H Y Y H R H P A Y T I L T V V S A A V A V L I G L T V A	wt sine2
356	I I H Y Y H R H P V Y T V I V L C G V A L A I L V G T A S S	Western equine
353	V V Y Y Y N R Y P L T T I I G L C T C V A I I M V S C V T S	Eastern equine
355	V Q Y Y Y G L Y P A A T V S A V V G M S L L A L I S I F A S	Semliki E2
356	I T H Y Y H R Y P M S T I L G L S I C A A I V T V S V A A S	venezuelan
354	V S H Y Y D L Y P Y W T I T V L A S L G L L I V I S S S G F S	Barmah
354	F S Y Y Y G L Y P A T T V A V C V G L A C V I L L A L S A S	middleburg
355	I T Y Y Y H S H P T T V V A C V T A A A V T L V M M C V G	ndumu

C W L C C K A R T K C L T P Y A L A P G A V V P T T L A L L Majority

400

410

420

386	A L C T C K A R R E C L T P Y A L A P N A V V P T S I A L L	wt sine2
386	A A C I A K A R R D C L T P Y A L A P N A T V P T A L A V L	Western equine
383	V W L L C R T R N L C I T P Y K L A P N A Q V P I L L A L L	Eastern equine
385	C Y M L V A A R S K C L T P Y A L T P G A A V P W T L G I L	Semliki E2
386	T W L F C K S R V S C L T P Y R L T P N A R M P L C L A V L	venezuelan
384	C F L C S V A R T K C L T P Y Q L A P G A Q L P T F I A L L	Barmah
384	C C L C V S A R N K C L T P Y A L T P G A V V P C T L S L L	middleburg
385	C S A C R V A R T R C L T P Y V L A P G S R V P L I L G L L	ndumu

C C A K S A R A

Majority

416	C C I R S A N A	wt sine2
416	C C I R P T N A	Western equine
413	C C I K P T R A	Eastern equine
415	C C A P R A H A	Semliki E2
416	C C A R T A R A	venezuelan
414	C C A K S A R A	Barmah
414	C C A P R A K A	middleburg
415	C C A K G A R A	ndumu

Decoration 'Decoration #1': Shade (with solid black) residues that match wt sine2 exactly.

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